Evidence on Developmental and Reproductive Toxicity of Methyl Isocyanate (MIC)

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October 21, 2010

Methyl Isocyanate (MIC)

- MIC is a highly reactive volatile and flammable chemical used to produce carbamate pesticides and other industrial chemicals
- Is a severe pulmonary irritant and is extremely toxic to humans after acute (short-term) exposure
- Effects of MIC on reproduction and development are based on exposures in Bhopal, India (1984) and animal studies conducted after the Bhopal disaster, in an attempt to understand the effects of this chemical

Pharmacokinetics

- Administration of ^{14}C-labeled MIC to laboratory animals (mouse, rat, guinea pig) by inhalation demonstrated that radioactivity rapidly distributed to brain, *lung, spleen, uterus*, placenta and *fetus*
- MIC-derived ^{14}C activity was detected in venous and arterial blood within minutes of exposure but it did not accumulate in the blood
- While majority of MIC is retained in the lungs, about 93-98% of absorbed MIC is eliminated in the urine within 3 days

Metabolism

Metabolites of MIC - methylamine, dimethylamine, trimethylamine and dimethylurea

In vitro data suggest that fetal toxicity of MIC

- Is not exerted through methylamines and is partly independent of maternal toxicity
- May result from the transfer of MIC across the placenta and interaction with fetal tissues

SMG, a conjugate of MIC and glutathione, exerted embryotoxic and dysmorphogenic effects in culture and may contribute to systemic toxicity of MIC

Non-DART effects

ACUTE EFFECTS:

- Pulmonary edema was the cause of death in most cases in Bhopal. Secondary respiratory infections, liver and kidney damage and eye problems noted in survivors of acute inhalation exposure with similar effects in animal studies
- LC50 levels in rodents following a 6 hour exposure were in the 6-12 ppm range

GENOTOXICITY & MUTAGENICITY:

- Results from *in vitro* studies indicate that MIC has the capacity to affect chromosome structure but not to induce gene mutation

CANCER:

- No information on the carcinogenic effects of MIC in humans is available and no tumors were significantly associated with MIC after animals were exposed once by inhalation

Studies in Animals

- Studies were conducted in both mice and rats
- No deaths among the adult mice at the doses administered, but the slope of the dose-response curve for MIC-induced toxicity is quite steep
- Exposures of mice to MIC at concentrations slightly higher than 3 ppm resulted in fatalities

Critical Differences Between Animal Studies and Human Exposure in Bhopal

- Some of the people in Bhopal exposed to much higher concentrations of MIC than those in the animal studies
- Animals exposed to pure MIC vapors while the people in Bhopal were exposed to MIC along with other reaction mixtures from the explosion

Developmental Toxicity Studies

Animal Studies

- Six studies reporting reproductive/developmental effects
- Two studies that did not report reproductive or developmental toxicity
- Additional related studies

Schwetz et al., 1987

Mating trial Study Design:

- 30 male and female Swiss (CD-1) mice/dose group mated following 4 consecutive days of exposure, 6 hours per day, to MIC vapors at 0, 1, or 3 ppm. Mating trials were conducted during *weeks 1, 8, and 17 following exposure.* The females were permitted to deliver their litters, and the pups were observed until 21 days of age.

Findings:

- No effect on body weight, demeanor, fertility, or litter size.

Schwetz et al., 1987

Perinatal Toxicity study design

Groups of 39-44 Swiss (CD-1) mice exposed to inhaled vapors of MIC at 0, 1, or 3 ppm, for 6 hours/day during GD14-17. Females were allowed to deliver.

Findings:

No effect on maternal survival, body weight, demeanor, or the length of gestation.

↑ in the number of dead pups at birth in both 1 and 3 ppm MIC groups.

↑ mortality among the neonates for these dose groups throughout lactation (i.e., a significant decrease in neonatal survival).

Varma et al., 1987

Study Design:

- Timed-pregnant mice exposed to MIC vapors for 3 hours

to (2 ppm, 6 ppm, 9 ppm or 15 ppm) on GD 8 or

to (9 ppm or 15 ppm) on GD 14.

- On GD 18 the animals were weighed and euthanized and the gravid uteri were exteriorized and fetuses removed by C-section

Varma et al., 1987

Findings:

- MIC vapor more toxic to the mother on GD 14 than GD 8

  (*all 3 animals exposed to 15 ppm for 3 hours on GD14 died within 24 hours compared to 2 of 18 i.e.,11% exposed on GD 8)*

- Exposure on GD 14 to MIC (9 ppm) caused higher mortality than exposure on GD 8

  (2 of 5 vs. 2 of 12)

Varma 1987

- Concentration-dependent ↓ in body weights of pregnant mice and relatively selective fetal toxicity
- Whole-body exposure of mice to 9 or 15 ppm MIC vapors for 3 hours on GD 8 led to resorption of implants (> 80%) with some maternal mortality
- ↑ in visceral abnormalities and ↓ in fetal and placental weights and fetal skeleton size
- ↓ 20% in the lengths of the mandible and bones of the extremities

Developmental effects after intraperitoneal (IP) exposure

- Fetal toxicity of MIC was produced after IP injections indicated that pulmonary irritation was not essential for the toxicity
- MIC exerts relatively selective fetal toxicity, although the mechanism remains to be identified
- No external malformations although some ↑ in visceral anomalies (thinning of myocardium in two fetuses, diaphragmatic hernia in two fetuses) noted

Singh et al., 1996

Study Design:

- Female rats exposed to 0, 0.212, 0.265 or 0.353 ppm of MIC vapors for half an hour and then mated with normal males of the same strain with standard teratology procedures on GD 20. Individual data not provided

Findings:

- Rate of resorptions ↑ in a dose-dependent manner
- Fetal weight ↓ by 3%, 4% and 7% in the low, high and highest dosed groups
- Teratological anomalies observed - wrist drop, everted claw, valgus deformity, syndactyly, blood clot formation, liver enlargement, cleft palate formation and unequal ribs

Developmental Toxicity
Other Relevant Information

Embryos exposed to MIC vapor both *in utero* or *in vitro* exhibited a concentration-dependent decrease in growth in culture

Exposure to MIC significantly decreased maternal plasma progesterone levels in mice that lost, but not in mice that retained, pregnancy

Authors concluded that fetal toxicity of MIC is partly independent of maternal toxicity and may result from the transfer of MIC across the placenta and interaction with fetal tissues

Developmental Toxicity
Other Relevant Information

Results suggest that the fetal toxicity of MIC is not exerted through methylamines the known metabolites of MIC, however in other cultured embryo experiments adverse effects were observed

Exposure of a conceptus *in utero* resulted in more toxicity than exposure of the gonadal cells prior to mating

Effects on Female Reproductive System

- MIC vapor resulted in ↓ placental weight
- Significant, dose-dependent ↑ in the number of implants absorbed
- While exposure to MIC significantly decreased maternal plasma progesterone levels in mice that lost, but not in mice that retained pregnancy, findings suggest ↓ in plasma progesterone was not the primary event leading to a loss of fetuses
- No adverse effects on reproduction were noted after exposure of female rats to MIC 70 days prior to mating

Effects on Male Reproductive System

- Transient ↓ in mating performance of MIC-exposed male mice cohabited with untreated females
- Loss of spermatozoa and degenerative changes in spermatocytes observed were reversible
- No effect on the incidence or distribution of resorptions in the pregnant females mated to the treated males
- No evidence of a dominant lethal effect in exposed male mice

Summary of Animal Data

Developmental Effects

- Animal data suggest an effect on fetal loss subsequent to *in utero* exposure
- Significant decrease in neonatal survival
- Adverse skeletal effects include a shortening of bones

Summary of Animal Data

Female Reproductive System

- ↓ placental weight
- Significant, dose-dependent ↑ in the number of implants absorbed

Male Reproductive System

- Reduction in mating performance and loss of spermatozoa was transient